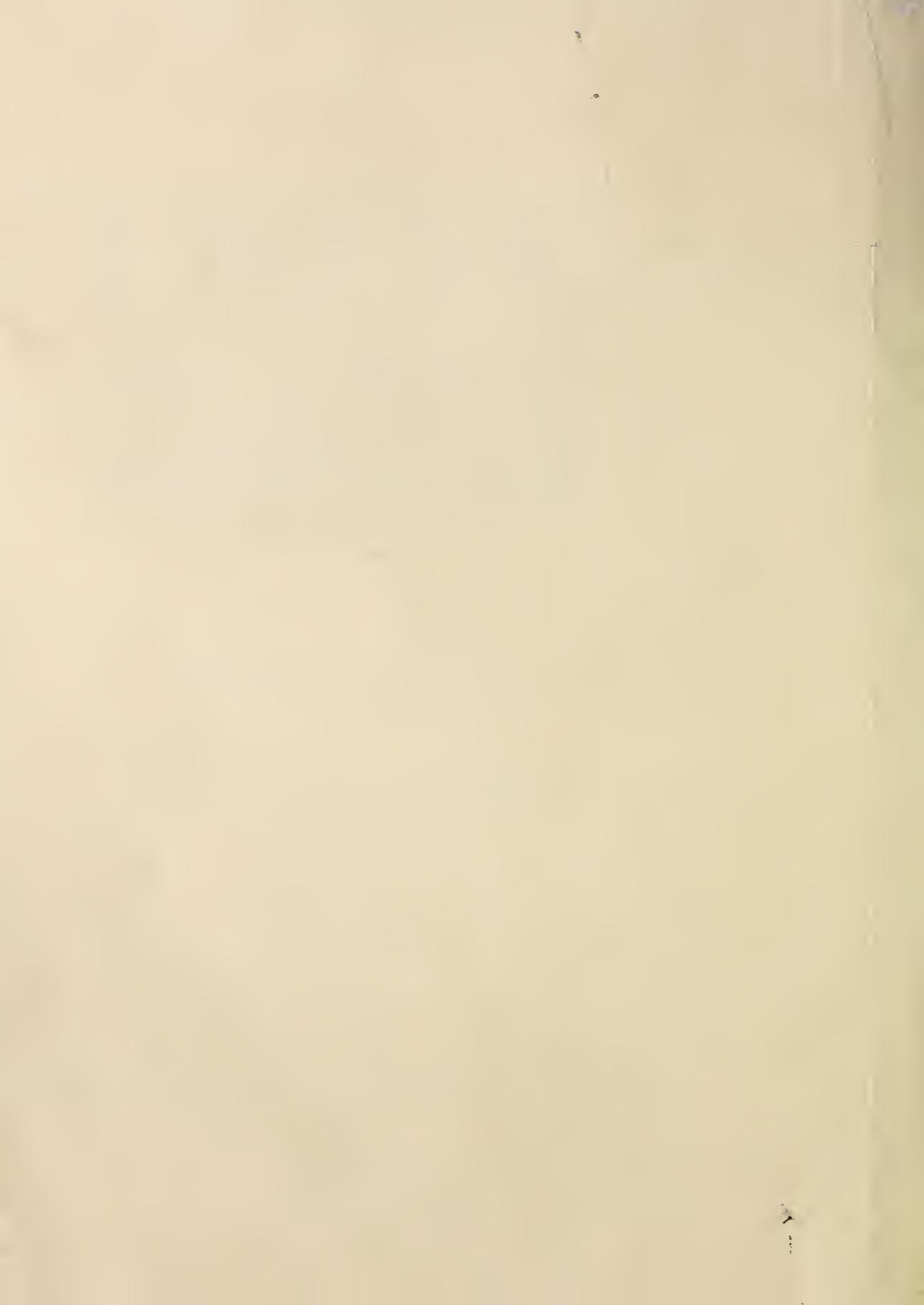


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## ENDRIN IN FOREST STREAMS AFTER AERIAL SEEDING WITH ENDRIN-COATED DOUGLAS-FIR SEED

by

Duane G. Moore, *Soil Scientist*  
Pacific Northwest Forest and Range Experiment Station  
USDA Forest Service Corvallis, Oregon

James D. Hall, *Associate Professor of Fisheries*  
Department of Fisheries and Wildlife  
Oregon State University, Corvallis, Oregon

Wayne L. Hug, *Aquatic Biologist*  
Lands Division, Oregon State Wildlife Commission  
Portland, Oregon



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## ABSTRACT

Extent and duration of endrin contamination in stream-water were determined after aerial seeding of two western Oregon watersheds with treated Douglas-fir seed. Detectable residues of endrin were found in a steep gradient stream for a period of less than 5 hours and in a slower flowing stream for 11 days. Endrin was again detected in the low gradient stream during the high flow of a winter storm 23 days after seeding. Maximum concentrations measured were well below reported 96-hour median tolerance limits for important fish species.

**Keywords:** Repellents (- animal damage control, water quality, endrin, seeding (direct), Douglas-fir, *Pseudotsuga menziesii*.

This publication reports research involving pesticides. It does not contain recommendations for their use, nor does it imply that the uses discussed here have been registered. All uses of pesticides must be registered by appropriate State and/or Federal agencies before they can be recommended.

**CAUTION:** Pesticides can be injurious to humans, domestic animals, desirable plants, and fish or other wildlife-- if they are not handled or applied properly. Use all pesticides selectively and carefully. Follow recommended practices for the disposal of surplus pesticides and pesticide containers.

This paper is also issued as Technical Paper No. 3709 of the Oregon Agricultural Experiment Station.

## INTRODUCTION

Endrin,<sup>1/</sup> a chlorinated hydrocarbon insecticide, has been extensively used in the Pacific Northwest for the past 15 years to protect directly sown Douglas-fir (*Pseudotsuga menziesii* [Mirb.] Franco) seed from seed-eating rodents. Seed is coated with endrin at a rate of 0.5 to 1 percent by weight (Radwan et al. 1970). Standard practice is to aerially sow treated seed at 0.56 to 1.12 kilograms per hectare.

Amounts of endrin thus introduced into the forest environment are very small (2.5 to 10 grams per hectare). However, endrin-treated seed may enter small streams flowing through seeded areas, and endrin is one of the most toxic pesticides to fish (Tarzwell 1963, Henderson et al. 1959, 1960). The 96-hour median tolerance limit (TL<sub>96</sub>) of coho salmon (*Oncorhynchus kisutch*) is 0.27 part per billion (p/b) endrin (Katz 1961, Katz and Chadwick 1961). Hence, there is concern about the potential hazard to aquatic organisms from the use of endrin-coated seed near forest streams.

Uptake of endrin and other chlorinated insecticides is very rapid (Johnson et al. 1971, Naqvi 1973, Wilkes and Weiss 1971), but Johnson et al. (1971) noted that degradation of the accumulated residues also starts during the same period. Biological magnification, stepwise food chain concentration, of endrin by aquatic invertebrates has not been reported. Bridges (1961) found that endrin applied to a pond disappeared rapidly from water, plants, bottom mud, and fish.

Rates of application and the total amounts of endrin introduced into forests by direct-seeding programs suggest that impact on the aquatic environment will be small, but firm knowledge of endrin residue levels is required. In the present study, we determined the extent and duration of endrin contamination in streamwater after aerial seeding of two western Oregon watersheds with treated Douglas-fir seed. Residue levels in aquatic insects, salmon eggs, and fish were monitored at one location.

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<sup>1/</sup> 1, 2, 3, 4, 10, 10-hexachloro-6, 7-epoxy-1, 4, 4a, 5, 6, 7, 8, 8a-octahydro-1, 4, 5, 8-endo-endo-dimethanonaphthalene.

## MATERIALS AND METHODS

### FIELD SAMPLING

Aerial seeding of two experimental watersheds in 1967 provided a unique opportunity to compare the extent and duration of streamwater contamination under different streamflow conditions. The watersheds are about the same size, and the length of stream channel covered by the seeding was similar (table 1). However, runoff characteristics differ greatly between the two streams, and these differences may influence maximum endrin concentrations measured and the length of time endrin can be detected in the water.

Watershed No. 1 at the H. J. Andrews Experimental Forest was aerially seeded on October 30, 1967, with 0.56 kilogram of 0.5-percent endrin-coated Douglas-fir seed per hectare. Needle Branch Watershed in the Alsea River Basin was seeded with 0.84 kilogram per hectare of 1.0-percent endrin-coated seed on December 18 and 26, 1967. The Needle Branch Watershed had been aerially seeded in January 1967 at this same rate (Marston et al. 1969); because of inadequate stocking with Douglas-fir, it was reseeded in December 1967.

Table 1.--Characteristics of two experimental watersheds in western Oregon: Watershed No. 1, H. J. Andrews Experimental Forest; and Needle Branch Watershed, Alsea Basin

Item	Watershed No. 1 <sup>1/</sup>	Needle Branch Watershed <sup>2/</sup>
Area (hectares)	96	71
Area seeded (hectares)	82	66
Aspect	WNW	SSW
Topography (average slope percent)	63	37
Elevation:		
Minimum (meters)	442	131
Maximum (meters)	1,013	366
Stream channel:		
Length (meters)	1,510	1,525
Elevation change (meters)	426	11

<sup>1/</sup> From Rothacher et al. (1967).

<sup>2/</sup> From Williams (1964).

Both experimental watersheds are equipped for streamflow measurements. Grab samples of water for endrin analysis were collected at the gaging stations before, during, and after seeding. Samples were taken in 4-liter metal containers, stored in ice, transported to the laboratory as soon as possible, and placed in a 2° C coldroom.

Water sampling for endrin residues began 30 minutes before seeding at each site and continued at 15-minute intervals for 2 hours after seeding started. The sampling interval then widened to one sample every 30 minutes for 1 hour, one 1-hour interval, and one sample every 2 hours for 4 hours, one 4-hour interval, one every 12 hours for 36 hours, one every 24 hours for 72 hours, and one sample at the end of the first week. Additional samples were taken at 2 weeks and 3 weeks after seeding and at 3-hour intervals during winter storms for 1 month.

A limited number of biological samples were collected at the Needle Branch site. About 1 week before seeding, eyed coho salmon eggs were obtained from the Fall Creek Salmon Hatchery and planted in separate chambers about 25 centimeters deep in the stream gravels to simulate the position of naturally deposited eggs. Small samples of two species of fish (coho salmon and the reticulate sculpin, *Cottus perplexus*) and several samples of aquatic insects were collected on several dates after seeding. The size and number of samples that could be collected were limited by the small size of the stream and the midwinter date of the seeding operation. Additional samples of coho salmon were obtained from Needle Branch and from adjacent untreated streams (Deer Creek and Flynn Creek) in January 1969, and samples of both fish and aquatic insects were obtained in July 1970. Aquatic biological samples were frozen as soon after collection as possible. Samples of the treated Douglas-fir seed were collected at the time of seeding and from 0.5-square-meter litterfall boxes about 1 month after seeding.

#### SAMPLE ANALYSIS

Water samples were analyzed for endrin by extracting the total volume collected (up to 4 liters) with *n*-hexane for 16 hours in a batch liquid-liquid partitioning extracting apparatus. Hexane extracts were dried with sodium sulfate, evaporated to 5 to 10 milliliters using a stream of nitrogen, and eluted through a 0.7-percent water-deactivated florisil column (Wood 1966). Eluate volumes were adjusted to 10 milliliters and analyzed by electron capture, gas-liquid chromatography (EC-GLC).

Biological samples were extracted by grinding with silica sand and 10 to 15 milliliters of hexane:isopropyl alcohol (4:1) in a glass mortar (volumes are for a 1- to 2-gram sample and are adjusted according to sample size). The solvent was decanted and filtered and the grinding repeated with

seven aliquots of fresh solvent. The residue and the filter paper were extracted with hexane in a soxhlet extractor for 20 hours. All extracts were combined, saponified, cooled, diluted with an equal volume of water, and extracted with hexane. The hexane extract was washed with water until the washings were neutral, dried with sodium sulfate, evaporated to 5 to 10 milliliters, and transferred to a 15-gram florisil column (Wood 1966). Columns were washed with 50 milliliters of benzene:hexane (30:70) followed by 75 milliliters of benzene:hexane (70:30) which eluted the endrin. The volume of eluate was adjusted with hexane for EC-GLC analysis.

A Microtek<sup>2/</sup> model GC-2000MF gas chromatograph with a 130-millicurie tritium electron capture detector and a 1.5-meter by 2-millimeter inside diameter glass column packed with 3-percent SE-30 on 80/100 mesh Gas Chrom Q was used for analysis of endrin. Inlet, column, and detector temperatures were 200°, 175°, and 195° C, respectively. Nitrogen carrier gas flowed at 40 milliliters per minute. Endrin was quantified by peak height, but data obtained were not corrected for recovery efficiencies. Average recoveries, based on analysis of spiked samples, were 97 percent for water samples, 94 percent for treated seed, and 92 percent for other biological material. Confirmation of the identity of endrin was by microcoulometric titration and by treatment with heat and hydrochloric acid to form the aldehyde and ketone isomers as reported by Phillips et al. (1962).

## RESULTS AND DISCUSSION

### ENDRIN RESIDUES IN STREAMWATER

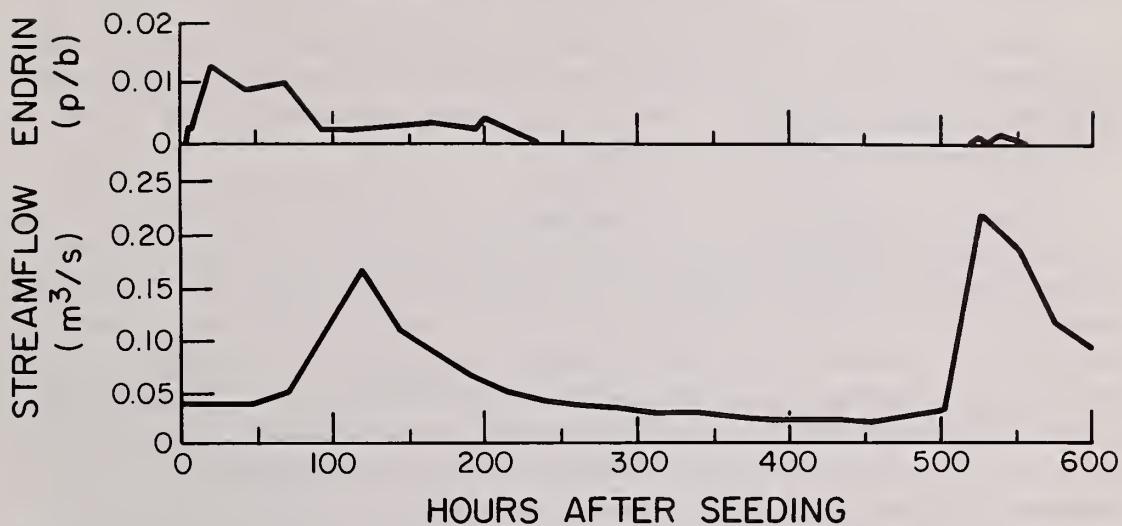
Instantaneous concentrations of endrin in samples from the two streams are shown in figure 1. The steep gradient of the stream channel in Watershed 1 resulted in the highest concentration of endrin (0.070 p/b) being measured 15 minutes after seeding started. Concentrations of endrin in the water changed rapidly, and the peaks coincide with the flight pattern of the helicopter. All detectable quantities of endrin in the stream were found in the first 5 hours after seeding began. Douglas-fir seed float, and seeds falling directly into the stream channel were carried downstream by the current. No seeds were found on the stream bottom.

In sharp contrast to Watershed No. 1, endrin was not detected in samples from Needle Branch until 6 hours after seeding started (fig. 1). The peak concentration of 0.013 p/b was reached in 21 hours, and residues remained near that level for 3 days. Concentrations then dropped to 0.002 to 0.004 p/b

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<sup>2/</sup> Mention of product or company does not imply endorsement by U.S. Department of Agriculture.

*NEEDLE BRANCH*  
*18 December 1967 - 12 January 1968*



*WATERSHED NO. 1*  
*30 October 1967*

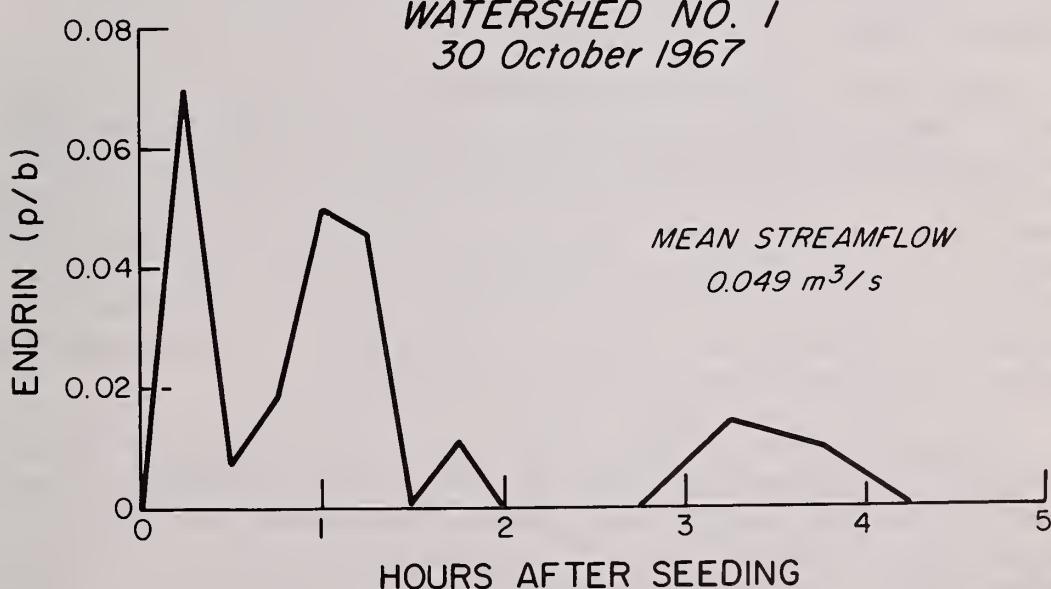


Figure 1.--Concentration of endrin in streamflow after aerial seeding with endrin-coated Douglas-fir seed: Needle Branch Watershed--seed treated with 1.0-percent endrin and sown at 0.84 kilograms per hectare; Watershed No. 1--seed treated at 0.5-percent endrin and sown at 0.56 kilograms per hectare.

for the next 5 days. Measurable amounts of endrin were again detected during the high flow of a winter freshet 3 weeks after seeding.

Half the treated seed was sown on December 18, and the other half on December 26, 1967. Unfortunately, too few samples were taken during the first several days after the second application to detect levels comparable to those measured earlier.

Residues were found in the Needle Branch stream during a winter freshet 3 weeks after seeding probably because of the relatively flat gradient of the stream channel. Endrin-treated seed falling into open water was not carried downstream as quickly as at Watershed No. 1, and some fir seed was undoubtedly trapped in small, quiet pools. Peak concentrations were lower at Needle Branch; one possible explanation is that, even though more time was available to dissolve endrin from the seed coating, more time was also available for adsorption of the residues on bottom and suspended sediments in the streams (Ferguson et al. 1965). Endrin leached from seed floating downstream was diluted by increased flow volumes in progressively larger streams, and concentrations in solution would be further decreased by adsorption on stream sediment.

The pattern of endrin concentration we found at Needle Branch is different from that reported by Marston et al. (1969) for a similar study of the same watershed a year earlier. They found endrin for only 2 hours after seeding started and again during a storm 6 days later. The maximum concentration found, 0.1 p/b, was in the second sample taken after seeding started. Stream discharge during that seeding operation, however, was about four times greater than in the present study. Higher streamflow reduced the influence of the flat stream gradient; thus their pattern of endrin outflow for Needle Branch closely resembles ours for Watershed No. 1.

Stream discharge data were used to calculate the total amount of endrin in runoff from each watershed (table 2). The endrin load was low and considerably less than the total amount applied directly to the stream channel. For example, the area of the stream channel in Watershed No. 1 is 0.14 hectare, and the amount of endrin applied to this surface area on treated fir seed was approximately 0.38 gram. Thus, the amount of endrin measured in the stream (0.011 gram) represents only about 3 percent of the potential load. Because of the steep gradient of this stream, we assume that the remainder of the applied endrin was carried from the watershed on floating seed. Amounts of endrin measured in Needle Branch samples were considerably higher because of the extended period of time over which detectable residues were present in the stream. Although stream contamination was extremely low, aquatic organisms were exposed to detectable residues for about 3 weeks.

Table 2.--Grams of endrin in streams of aerially seeded watersheds in western Oregon based on water samples collected between January 23, 1967, and January 10, 1968

Experimental watershed	Total applied to watershed	Total recovered in stream	Total in stream as percent of total applied to watershed
-----Grams----- -----Percent-----			
Watershed No. 1, H. J. Andrews Experimental Forest <sup>1/</sup>	230	0.011	0.005
Needle Branch Watershed, Alsea Basin: <sup>2/</sup>			
January 1967 <sup>3/</sup>	558	.697	.125
December 1967	336	.190	.057

<sup>1/</sup> Douglas-fir seed treated with 0.5-percent endrin and sown at 0.56 kilogram per hectare.

<sup>2/</sup> Douglas-fir seed treated with 0.6-percent endrin and sown at 0.84 kilogram per hectare.

<sup>3/</sup> Calculated from data of Marston et al. (1969).

#### ENDRIN RESIDUES IN BIOLOGICAL SAMPLES

Data are not presented for samples of aquatic insects, fish, and planted salmon eggs collected before and during the first 4 weeks after aerial seeding of the Needle Branch Watershed. These samples had been frozen as collected and were all processed through the laboratory for residue analysis at the same time. Several, and perhaps all, of these samples were apparently contaminated before or during analysis. All pretreatment samples contained otherwise unexplainable endrin residues, since it is doubtful that any residues from the January 1967 seeding would have persisted through the spring and fall storms. Posttreatment data were extremely variable and exhibited unexplained temporal relationships.

Although inadvertent contamination of these biological samples makes it impossible to draw specific conclusions, a few general observations can be made. Endrin residues were present in all biological materials sampled. In

general, residue concentrations increased and then gradually declined after seeding, but it is not possible to quantify actual maximum residue levels resulting from treatment. Even with sample contamination, however, maximum levels of endrin residues in fish did not reach 100 p/b. Residue levels well above 100 p/b endrin did not result in mortality of steelhead fingerlings in a live-box test conducted in California (California Department of Fish and Game 1967).

Additional biological samples were collected 12 and 30 months after treatment to determine whether any detectable residues had persisted from the application of endrin-coated Douglas-fir seed in December 1967. Wild adult coho salmon were obtained from Needle Branch and Deer Creek, and a sample of retained eggs was taken from the Deer Creek fish in January 1969. Coho fingerlings and aquatic insects were collected from Needle Branch and Flynn Creek in July 1970. No endrin was detected in any of these samples.

#### POSSIBLE HAZARD TO AQUATIC ENVIRONMENT

Cope (1966) and Hunt (1966) conducted laboratory bioassay tests to determine the toxicity of endrin-treated conifer seed to fish and wildlife. Two endrin-treated Monterey pine seeds (0.655-percent endrin) in a 20-liter aquarium were fatal to rainbow trout. Douglas-fir seed containing 200 micrograms endrin per seed caused complete mortality of five rainbow trout in 1 day when 21 seeds were added to 15 liters of aquarium water. Endrin leached from six seeds caused complete mortality in 3 days.

Field studies, however, conducted in California and Oregon have indicated that application of endrin-coated conifer seeds to Pacific slope watersheds may not pose a serious hazard to aquatic organisms (Morton 1967, California Department of Fish and Game 1967). In bioassay tests, fingerling steelhead were placed in live-cars in a flowing stream, and treated pine seed was put in cheesecloth bags and placed in the stream above each live-car. A safety factor for fish was demonstrated of greater than 26 times the amount of endrin that would fall into the stream at the prescribed application rate of 0.84 kilogram per hectare (California Department of Fish and Game 1967). Other studies carried out cooperatively by the Bureau of Commercial Fisheries and the California Department of Fish and Game (1967) indicate that endrin residues are not accumulating in shellfish in bays and estuaries of central and northern California.

## CONCLUSIONS

Aerial application of endrin-coated Douglas-fir to reforest harvested or burned watersheds as previously practiced in the Pacific Northwest does not appear to constitute a serious hazard to aquatic habitats. Although rates of seed treatment and dissemination are very low, endrin concentrations known to be toxic to several important species of anadromous fish can result if treated seed fall directly into open ponded water. Static water bioassay tests show that treated fir seed can release sufficient endrin to cause mortality. However, adsorption of endrin residues on stream sediments and continuous dilution in rapidly moving waters of forest streams prevent the development of hazardous exposure conditions.

Data obtained in the present study indicate that direct seeding does result in detectable residues of endrin in streamwater and that the level and duration of contamination depend on the profile of the stream channel. Aquatic organisms normally will not be exposed to chemical residues for more than a few hours in streams with steep gradients or more than several days in slower flowing streams. The maximum concentrations measured are well below reported 96-hour median tolerance limits for important fish species. Detectable levels of endrin were found in the Needle Branch stream up to 23 days after seeding, probably because high flow during a winter storm picked up additional treated seed from along the stream banks. However, actual exposure of fish and aquatic insects amounted to only 3 days to 0.010 p/b endrin and another 8 days to an average of 0.002 p/b.

Endrin residues were found in all biological samples collected during the first month after seeding. Unfortunately, many of these samples were contaminated during analysis, but maximum residue levels in fish were still less than 100 p/b endrin. In general, residues in the aquatic organisms began to dissipate as soon as detectable levels disappeared from the surrounding water. Similar results have been reported by Bridges (1961) and Ferguson et al. (1966).

Although endrin applied at high rates can persist in soils for many years (Nash and Woolson 1967), residues did not appear to accumulate or persist in the aquatic ecosystems of the treated watersheds. Stream bottom sediments were not sampled, but these streams are subject to frequent flushing during the high flow of winter storms, and it is reasonable to assume that adsorbed endrin would soon be carried downstream. Biological samples collected from treated and untreated streams 12 and 30 months after treatment contained no endrin residues. Stream contamination which did occur in the present study, even though small, might be minimized or eliminated by leaving appropriate buffer strips and avoiding direct application to open water and streambanks.

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